



Effect of alpine plant species on *in vitro* ruminal fermentation characteristics

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Introduction

Ruminal
fermentation



Methane
production



3.3% to the total
greenhouse gas emissions

(Knapp et al. 2014)

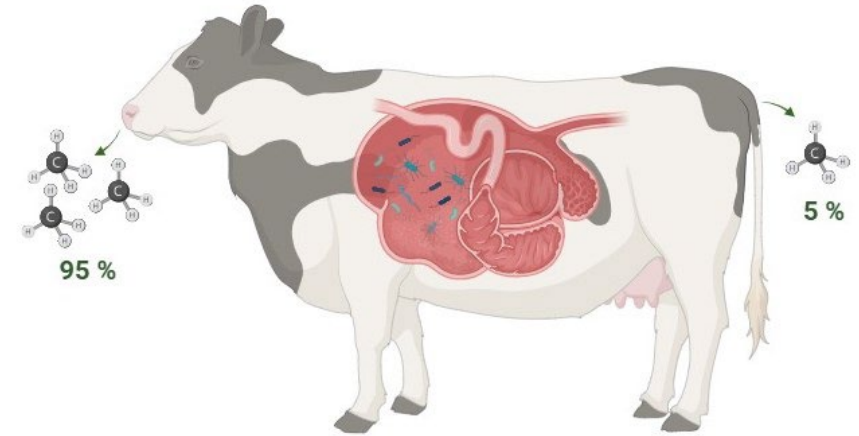


significant **inefficiency** in
feed utilization

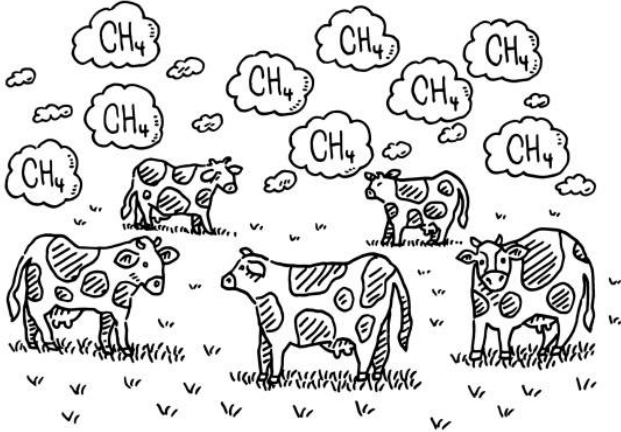


gross energy loss of
around **2 to 12%**

(Johnson & Johnson 1995)



Introduction



To reduce CH_4 emissions

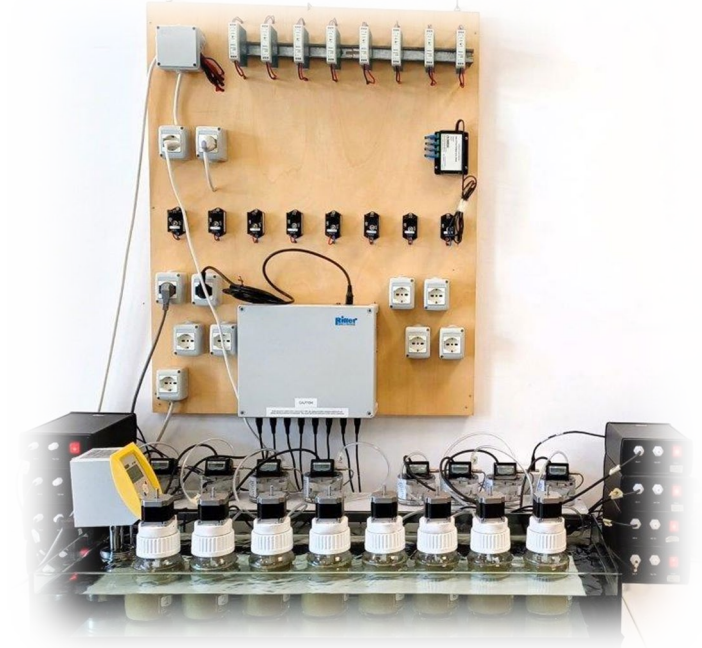


Diet management



effect of different **plant species** on:
methane production
ruminal fermentation parameters

in vitro system for continuous
measurement



Materials And Methods – Plant sampling



11 plant species



grazing dairy cows'
diets

sampled at the start of
the **grazing season**

1. availability
2. palatability



plant was cut about **3 cm**
above the ground



0.5 kg of fresh matter
was collected for each



vacuum packed stored at
refrigeration temperature
(**4°C**)

Materials And Methods - Plants

Achillea millefolium
Common yarrow



Carum carvi
Caraway



Festuca rubra
Red fescue



Hypericum maculatum
Imp. St John's wort



Lotus corniculatus
Bird's-foot trefoil



Poa alpina
Alpine bluegrass



Potentilla erecta
Tormentil



Prunella grandiflora
Large selfheal



Ranunculus acris
Buttercup



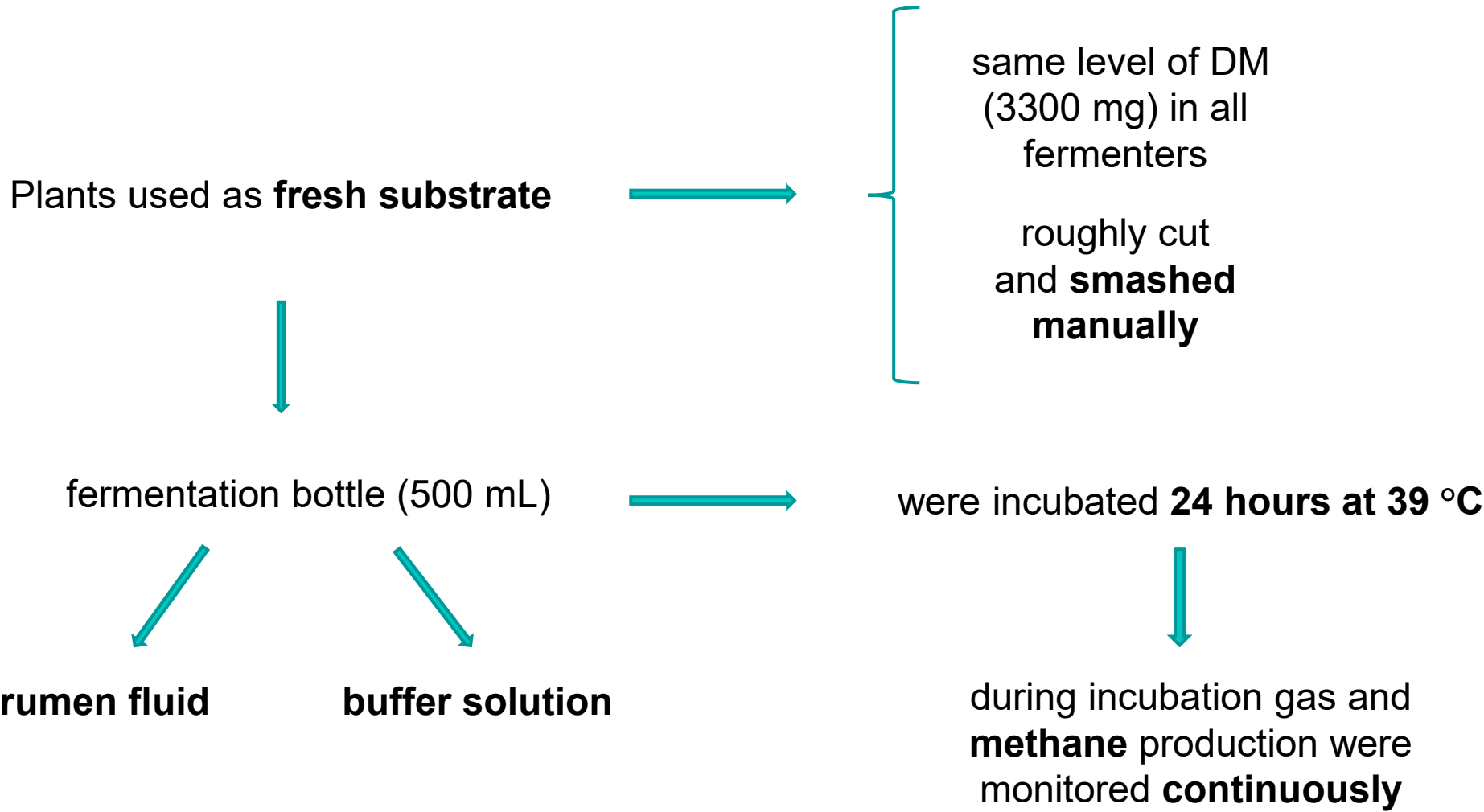
Trifolium repens
White clover



Veronica chamaedrys
Germander speedwell



Materials And Methods – *In vitro* trial



Statistical Analysis

**Balanced incomplete block
design**

(Cox and Reid, 2000)



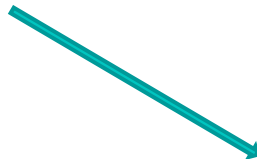
11 fermentation runs

five distinct fermentation runs (each plant)

Gompertz model



percentage of methane in total gas
recorded **each hour** from **each**
fermentation bottle



$$Y_t = B(\exp(-C\exp^{-At}))$$

Proximal composition of plants used in the *in vitro* trial



L. corniculatus



T. repens



F. rubra



P. alpina

	Composition (% DM)						
	DM	CP	NDF	ADF	ADL	Ash	EE
Achillea millefolium	25.7	16.8	49.2	26.4	8.06	7.73	2.02
Carum carvi	18.5	16.1	41.9	24.5	6.15	9.29	1.72
Festuca rubra	40.3	7.59	74.8	40.7	5.77	2.53	1.50
Hypericum maculatum	31.7	11.5	56.2	38.4	21.7	5.57	1.61
Lotus corniculatus	22.5	20.3	42.6	24.9	10.4	7.37	1.92
Poa alpina	43.7	6.92	69.1	34.7	4.48	2.78	1.96
Potentilla erecta	32.2	12.0	51.6	28.8	8.70	5.20	1.41
Prunella grandiflora	20.5	10.6	42.8	24.0	10.2	8.61	1.22
Ranunculus acris	23.0	8.90	51.7	32.1	8.00	5.83	2.18
Trifolium repens	23.0	24.6	35.9	19.9	6.16	10.7	1.85
Veronica chamaedrys	28.0	11.4	44.5	27.3	11.7	6.28	0.76

Results and Discussion

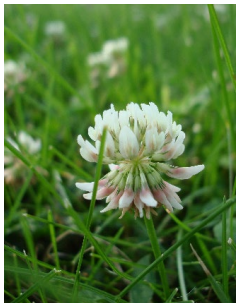
Fermentative traits obtained after 24 hours of *in vitro* fermentation



H. maculatum



P. alpina



T. repens



P. grandiflora

	Total VFA (mM)	% Total VFA			C2:C3	NH3 (mg/dL)
		C2	C3	C4		
Achillea millefolium	36.4	71.4 ^{abc}	12.9 ^{abc}	11.3 ^{ab}	5.57 ^{ab}	60.9 ^{ab}
Carum carvi	36.3	72.0 ^{abc}	13.7 ^{ab}	10.3 ^{ab}	5.36 ^b	55.4 ^{abc}
Festuca rubra	38.6	70.0 ^{bc}	13.2 ^{ab}	12.3 ^{ab}	5.37 ^b	55.6 ^{abc}
Hypericum maculatum	26.3	74.4 ^a	11.2 ^c	10.5 ^{ab}	6.75 ^a	53.1 ^{abc}
Lotus corniculatus	29.5	70.8 ^{abc}	13.5 ^{ab}	12.1 ^{ab}	5.34 ^b	56.5 ^{ab}
Poa alpina	34.6	69.2 ^c	14.0 ^a	12.3 ^{ab}	4.93 ^b	60.2 ^{ab}
Potentilla erecta	32.8	72.6 ^{abc}	12.0 ^{bc}	11.3 ^{ab}	6.10 ^{ab}	50.4 ^{bc}
Prunella grandiflora	28.4	71.6 ^{abc}	12.2 ^{abc}	13.0 ^a	5.97 ^{ab}	42.5 ^c
Ranunculus acris	34.8	73.3 ^{ab}	12.3 ^{abc}	10.6 ^{ab}	5.97 ^{ab}	55.0 ^{abc}
Trifolium repens	50.4	73.9 ^a	12.6 ^{abc}	9.43 ^b	6.17 ^{ab}	65.5 ^a
Veronica chamaedrys	35.9	71.8 ^{abc}	13.1 ^{bc}	11.3 ^{ab}	5.48 ^{ab}	50.3 ^{bc}
Significance	NS	**	**	**	**	**
RMSE	11.5	1.57	0.77	1.33	0.56	5.98

Gas and methane values after 24 hours of *in vitro* fermentation



C. carvi



P. erecta



F. rubra



R. acris

	Gas (mL/g DMD)	Methane (mL/g DMD)	Methane (%)
Achillea millefolium	432 ^{ab}	106 ^{ab}	27.8 ^{ab}
Carum carvi	523 ^a	126 ^a	24.5 ^{ab}
Festuca rubra	398 ^{abc}	112 ^a	28.9 ^a
Hypericum maculatum	254 ^{bc}	64.6 ^{ab}	25.3 ^{ab}
Lotus corniculatus	319 ^{abc}	80.7 ^{ab}	24.9 ^{ab}
Poa alpina	339 ^{abc}	89.2 ^{ab}	26.7 ^{ab}
Potentilla erecta	191 ^c	49.2 ^b	24.6 ^{ab}
Prunella grandiflora	303 ^{abc}	79.8 ^{ab}	26.6 ^{ab}
Ranunculus acris	513 ^a	122 ^a	24.2 ^b
Trifolium repens	253 ^{bc}	65.3 ^{ab}	26.6 ^{ab}
Veronica chamaedrys	312 ^{abc}	73.7 ^{ab}	24.3 ^{ab}
Significance	**	**	*
RMSE	103	26.8	1.99

Kinetics parameters

- **A** is a constant that describes the decline of the production rate
- **B** is the asymptotic methane volume
- **C** is the specific CH₄ production rate dependent on time
- **TMFR** is the time to maximum fermentation rate
- **MFR** is the maximum fermentation rate
- **Lag** is the lag phase.

	A	B, %	C, 1/h	TMFR, h	MFR, %/h	Lag, h
Achillea millefolium	0.82 ^{bcd}	25.0 ^{bc}	4.57	2.22 ^{cde}	7.62 ^{ab}	0.72
Carum carvi	1.02 ^a	24.5 ^c	4.87	1.87 ^e	8.88 ^a	0.60
Festuca rubra	0.38 ^f	29.1 ^a	4.07	3.57 ^a	4.00 ^d	0.84
Hypericum maculatum	0.49 ^{ef}	23.9 ^c	2.98	2.64 ^{bc}	4.58 ^{cd}	0.37
Lotus corniculatus	0.75 ^{cd}	24.9 ^c	4.73	2.26 ^{cde}	6.89 ^{abc}	0.73
Poa alpina	0.43 ^f	28.8 ^a	4.30	2.88 ^b	4.89 ^{cd}	0.73
Potentilla erecta	0.37 ^f	26.1 ^{abc}	2.62	2.31 ^{bcde}	3.51 ^d	0.20
Prunella grandiflora	0.91 ^{abc}	28.3 ^a	5.65	2.43 ^{bcde}	9.30 ^a	0.90
Ranunculus acris	0.95 ^{ab}	24.2 ^c	4.87	1.88 ^e	8.27 ^{ab}	0.67
Trifolium repens	0.63 ^{de}	27.8 ^{ab}	4.24	2.46 ^{bcd}	6.49 ^{bc}	0.76
Veronica chamaedrys	0.87 ^{abc}	24.1 ^c	5.61	1.99 ^{de}	7.91 ^{ab}	0.61
Significance	**	**	NS	**	**	NS
RMSE	0.12	1.73	1.10	0.36	1.40	0.28

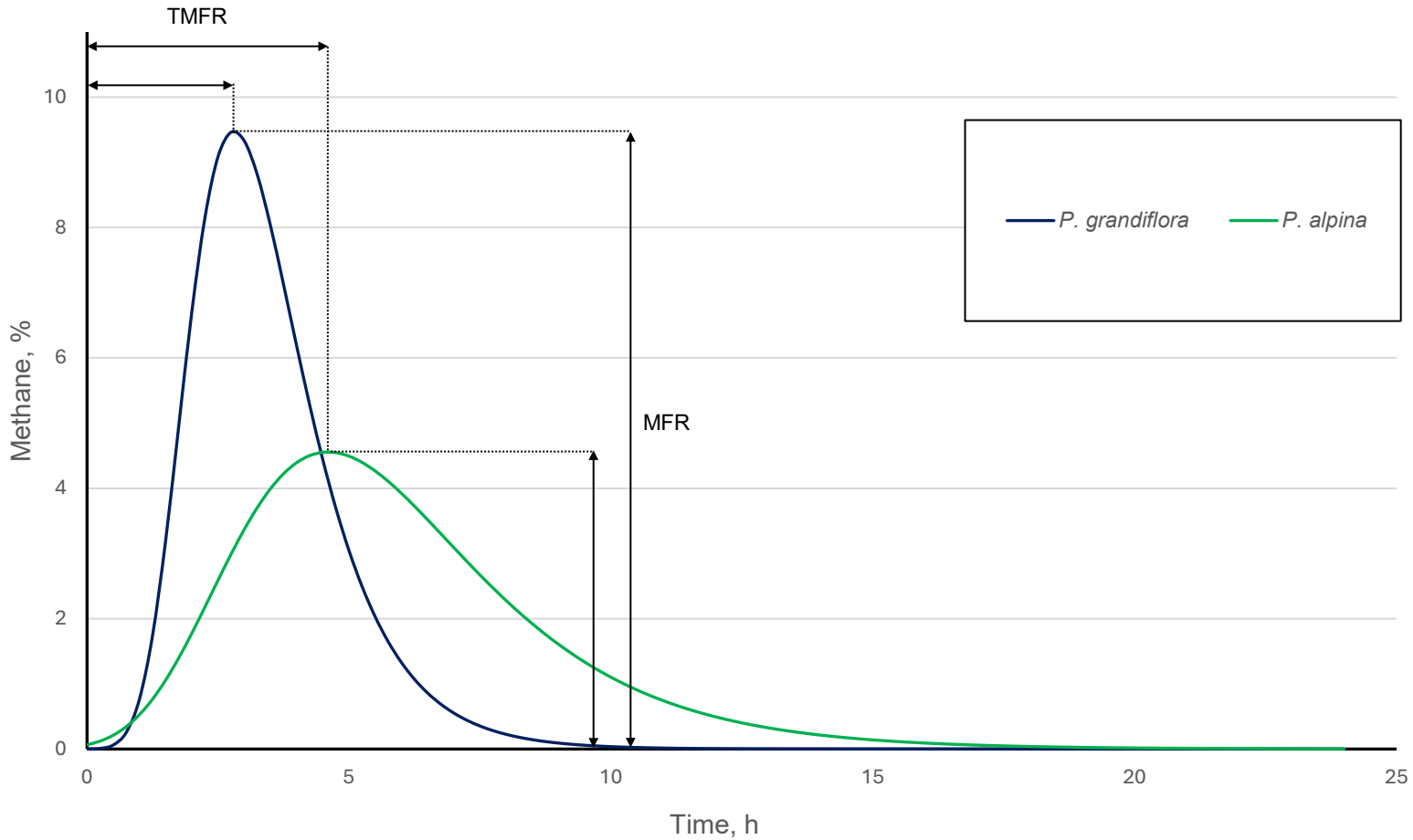
Results and Discussion

Methane yield

	CH4 (%)	A	C (1/h)	TMFR (h)	MFR (%/h)	Lag (h)
Poa alpina	26,7	0,43	4,30	2,88	4,89	0,73
Prunella grandiflora	26,6	0,91	5,65	2,43	9,30	0,90

- Prunella grandiflora: intense but short-lived production of CH4

-Poa alpina: prolonged CH4 production over the time but less intense



Conclusion

In vitro gas production systems are widely used to rapidly detect methane resulting from the ruminal fermentation of several substrates, including plant species

The chemical composition of plants influences fermentation

None of the examined plants drastically reduce CH₄

The study of kinetics allows for the differentiation of plants that only appear to be similar (after 24 h of fermentation)

Acknowledgements

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***Thanks for
your kind attention!***